Another PDT modality in the prior art teaches the destruction of abnormal cells that are circulating in the blood using light therapy, while leaving the blood vessels intact (see, for example: U.S. Patent No. 5,736,563, Richter *et al.*; WO 94/06424, Richter; WO 93/00005, Chapman *et al.*; U.S. Patent No. 5,484,803, Richter *et al.*, and WO 93/24127, North *et al.* Instead, it might be preferable to deliberately damage and occlude blood vessels that form the vasculature supplying nutrients and oxygen to a tumor mass, thus rendering a given volume of abnormal tissue in the tumor (not circulating cells) ischemic and anoxic and thus promoting the death of the tumor tissue serviced by these blood vessels.

Please replace the paragraph on page 6, lines 17-26, with the following paragraph:

PDT of locally recurrent breast cancer (LRBC) with lutetium texaphyrin has been reported by T.J. Wieman *et al.*, in program/proceedings, *American Society of Clinical Oncology*, Vol. 18, P. 111A (1999). This study by Wieman *et al.* involved the treatment of superficial recurrent chest wall breast cancer. Lutrin™ (lutetium texaphyrin, brand; Pharmacyclics, Inc, Sunnyvle, CA) was administered by injection at a dose of 1.5 mg/Kg to 4.0 mg/Kg and followed by chest wall illumination by 150 joules or 100 joules of light at 732 nm using laser or LED device. However, this study did not suggest or disclose the use of transcutaneous light delivery to treat a subcutaneous tumor mass. Further, at the light dosage employed, at sustained delivery of light at the reported intensity may not be possible without adverse reactions.

Please replace the paragraph on page 6, line 27 through page 7, line 4, with the following paragraph:

It is apparent that the usual method of administering PDT to treat bulky tumors, which relies on invasive introduction of optical fibers, is not the best approach. It would be highly advantageous to apply light transcutaneously in a completely noninvasive method to treat such large tumors (as well as small and even microscopic tumors), without risking damage to non-target tissues, such as skin and normal subcutaneous tissue. Instead of the conventional technique, a

method of photoactivation and a series of photosensitizer constructs is needed that enable PDT induced cytotoxicity, on both a macro and microscopic scale, without risk to the cutaneous layer, or any surrounding normal tissues. Also, the therapeutic index should be enhanced if a specific photosensitizer drug targeting scheme is employed.

Please replace the paragraph on page 8, lines 13-25, with the following paragraph:

In one application of the invention, the target tissue is vascular endothelial tissue. In another application, the target tissue is an abnormal vascular wall of a tumor. As further defined, the target tissue is selected from the group consisting of: vascular endothelial tissue, an abnormal vascular wall of a tumor, a solid tumor, a tumor of a head, a tumor of a neck, a tumor of a gastrointestinal tract, a tumor of a liver, a tumor of a breast, a tumor of a prostate, a tumor of a lung, a nonsolid tumor, malignant cells of one of a hematopoietic tissue and a lymphoid tissue, lesions in a vascular system, a diseased bone marrow, and diseased cells in which the disease is one of an autoimmune and an inflammatory disease. In yet a further application of the present invention, the target tissue is a lesion in a vascular system. It is contemplated that the target tissue is a lesion of a type selected from the group consisting of atherosclerotic lesions, arteriovenous malformations, aneurysms, and venous lesions.

Please replace the paragraph on page 9, lines 11-16, with the following paragraph:

Preferably, photosensitizing agents or prodrugs are of a chemical composition that allows them to cross fenestrations and gaps in tumor vessels and bind to the ablumenal as well as the lumenal side of the blood vessels. As target cell (e.g., a selected tumor cell type) cytotoxicity occurs adjacent to blood vessels, cell swelling and release of cellular contents leads to further inflammation which augments the occlusion process from the ablumenal side.

Please replace the paragraph on page 13, lines 25-33, with the following paragraph:

FIGURE 1 illustrates transcutaneous delivery of light 12 from an external source 10 to a relatively deep tumor 14, or to a plurality of small, but relatively deep tumors 16. The light emitted by external source 10 is preferably of a longer waveband, but still within an absorption waveband of the photosensitive agent (not shown in this Figure) that has been selectively linked to tumor 14 and smaller tumors 16. The longer wavelength of light 12 enables it to pass through a dermal layer 18 and penetrate into the patient's body beyond the depth of tumor(s) being treated with targeted PDT. In these two examples, the PDT is directed specifically at target cells in tumor 14 or in tumors 16.

Please replace the paragraph on page 17, lines 5-14 with the following paragraph:

"Radiation" as used herein includes all wavelengths and wavebands. Preferably, the radiation wavelength or waveband is selected to correspond with or at least overlap the wavelength(s) or wavebands that excite the photosensitive compound. Photosensitive agents or compound typically have one or more absorption wavebands that excite them to produce the substances, which damage or destroy target tissue, target cells, or target compositions. Even more preferably, the radiation wavelength or waveband matches the excitation wavelength or waveband of the photosensitive compound and has low absorption by the non-target cells and the rest of the intact animal, including blood proteins. For example, a preferred wavelength of light for ICG is in the range 750-850 nm.

Please replace the paragraph on page 26, lines 16-20, with the following paragraph:

The fluence rate employed in this Example represented about 150-180 mW/cm^2, with a total fluence more than 20,000 Joules. The preferable fluence rate contemplated more broadly by the present invention is between about 5 mW/cm^2 and about 100 mW/cm^2, more preferably, between about 10 mW/cm^2

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and about 75 mW/cm^2, and most preferably, between about 15 mW/cm^2 and about 50 mW/cm^2.

Please replace the paragraph on page 31, line 22, through page 32, line 18, with the following paragraph:

In this Example, a capsular or pill-shaped and sized light source 120 is administered orally to a patient, so that it passes into the stomach 118 of the patient, where it administers light 122. Alternatively, an optical fiber (not shown) may be passed into the stomach via the nasopharynx to administer light 122 to the treatment site. In order to implement targeted PDT for treating ulcers in humans, and APC 124, with antibody 131 is targeted against a suitable *Helicobacter pylori* antigen 126. The APC is formulated into an ingestable compound that releases the APC to a gastric mucus/epithelial layer 128 where the bacterium is found. The APC is ingested at a time when the stomach and duodenum is substantially empty in order to promote binding of the APC to bacterium 130. Any unbound APC is diluted by gastric juice and carried distally by peristalsis to be eliminated from the body in fecal matter. Light sources suitable for intraluminal passage are disclosed in any one of U.S. Patent Nos.: 5,766,234; 5,782,896; 5,800,478; and 5,827,186, the disclosure of each being specifically hereby incorporated herein in its entirety. Alternatively, light source 120 in capsule or pill form, e.g., as disclosed in copending commonly assigned U.S. Patent application Serial No. 09/260,923, entitled, "Polymer Battery for Internal Light Device," filed on March 2, 1999 and which is hereby incorporated in its entirety by reference herein, is used for activating the APC. The light source is preferably energized just prior to its ingestion or remotely after ingestion, when in the stomach or in a desired intraluminal passage. If necessary, multiple light sources are ingested to insure that adequate photoactivation of the localized APC occurs sufficient to kill the bacterium. Light is delivered at a relatively low fluence rate but at a high total fluence dose, as discussed above. The light source(s) may be deactivated after passage beyond the duodenum to avoid unwanted distal photoactivation. In

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this manner, a photosensitizing agent 132 comprising the APC is activated topically without the need for a procedure such as endoscopy with fiberoptic gastric illumination in order to provide the activating light. Since the APC is targeted, nonspecific uptake by normal tissue and other normal compositions of the body is minimized in order to prevent injury to normal gastric tissue and problems with the gastric system.

Please replace the paragraph on page 34, lines 12-25, with the following paragraph:

A photosensitizer conjugate is formulated which binds with great affinity to *Streptococcus pneumoniae* and *Hemophilus influenzae* in a selective manner. The APC is formulated into an injectable compound, which can be administered intravenously or instilled topically into the middle ear via a previously placed tympanostomy tube. The drug is activated using light emitted by a small light source about the size, shape, and weight of a hearing aid, which is disposed behind the ear and aimed at the middle ear, so that the light passes into the middle ear transcutaneously.

Step 1 the APC fluid formulation is instilled into the middle ear.

Step 2 Sufficient time is allowed to elapse to allow binding of the APC with the disease organisms, and then, any excess fluid is drained away by gravity or actively aspirated using a needle and syringe.

Step 3 The light source is positioned behind the ear and activated. The light source need not be very intense since the middle ear cavity is very small. Further, The fluence rate and total fluence dose may be followed as discussed above.

Please replace the paragraph on page 35, lines 14-29, with the following paragraph:

This Example uses the present invention for the treatment of an organ infiltrated with tumor tissue. Reference is made to FIGURE 9. Specifically, light 140 is administered by transillumination through liver tissue 148 from an implanted light source 144 that is disposed external to the surface of liver 142,

but within the patient's body underneath the skin layer (18). In this embodiment, a patient is injected intravenously with a photosensitizer agent ICG, conjugated to an antibody that is specific to vascular endothelial antigen (not separately shown) on a tumor 146, so that the antibody binds with the antigen, but not to other tissue in the liver. The optimal dose of ICG will be empirically determined, for example, via a dose escalation clinical trial as is so often performed to evaluate chemotherapeutic agents. One or more light source probes 144 are surgically implanted (e.g., endoscopically) adjacent to, but not invading parenchymal tissue 148 of liver 142. After delaying a time sufficient to permit clearing of the photosensitizer conjugate from the non-target tissues, the light source(s) is(are) activated, irradiating the target tissue with light 140 at a relatively low fluence rate, but administering a high total fluence dose of light in the waveband from about 750 nm to about 850 nm.

Please replace the paragraph on page 36, lines 7-13, with the following paragraph:

The present example employs Lutrin™ (lutetium texaphyrin, brand: Pharmacyclics, Inc, Sunnyvale, CA) as a photosensitizer drug compound. A proportion of Lutrin™ 150 begins to clear from normal tissue 144 in about 3 hours, a larger proportion clears from normal tissue in about 8 hours, with an even greater proportion clearing in about 16 hours. The predominant amount of photosensitizer clears from normal tissue in about 24 hours from administration of the agent. However, tumor tissue 146 retains the photosensitizer up to 48 to 96 hours after administration.

Please replace the paragraph on page 37, lines 10-17, with the following paragraph:

Example 11

PDT of Human Gall Bladder Carcinoma Cells - *In Vitro*

Human gall bladder carcinoma cells are grown to confluence in 12-well plates. An array of light emitting diodes are suspended above the plates to provide illumination. The cells are loaded with a variety of photosensitizers and

illuminated for prolonged periods of time ranging from 48 - 72 hours with only 30-85 microwatts (μW) of light in some cases. In all cases virtually all tumor cells are reliably killed and histologically exhibit irreversible changes leading to cell death. (See Figures 11-14)

Please replace the paragraph on page 37, lines 19-24, with the following paragraph:

Example 12

PDT of Human Gall Bladder Carcinoma Cells - *In Vivo*

A series of experiments were performed using nude mice growing transplanted human tumors. The mice are injected with various photosensitizers and the tumors illuminated with low fluence of only 30 μW of light over a 72 hour time period. Extended tumor necrosis was observed.

Please replace the paragraph on page 37, line 30, through page 38, line 4, with the following paragraph:

Two experimental mice were injected with epithelial cancer cells preincubated with 10 micrograms of Pheophorbide A. These mice were exposed to 660 nm (peak) light for about 48 hours (30 microwatts per cm^2) with no tumor growth after 1.5 months. The control animals ("dark controls") maintained in the absence of light developed a large tumor. Another two mice with established tumors were injected with 50-100 micrograms of Pheophorbide A into the lesion and exposed to 660 nm light (30 microwatts per cm^2) for 72 hours. Extensive tumor necrosis resulted after 7 days, but no effect was observed in the dark control animals.

Please replace the paragraph on page 38, lines 7-12, with the following paragraph:

Two experimental mice were injected with epithelial cancer cells preincubated with 20 micrograms of Chlorin e6. These mice were exposed to 660 nm light for about 48 hours (30 microwatts per cm^2) with no tumor growth after 1.5 months. The dark control developed a large tumor. Another two mice were injected with 100-150 micrograms of Chlorin e6 intratumorally and then

exposed to 660 nm light (30 microwatts per cm^2) for 72 hours. Extensive tumor necrosis resulted in both after 7 days.

Please replace the paragraph on page 38, lines 15-19, with the following paragraph:

Five experimental mice bearing established tumors were injected with 1 mg Hpd intraperitoneally followed by exposure to 630 nm (peak) light (30 microwatts per cm^2) for 72 hours. Extensive tumor necrosis was seen upon gross and histological examination in all cases after 7 days. There was no effect observed on control animals maintained in the absence of light (dark control mice).

Please replace the paragraph on page 38, lines 25-32, with the following paragraph:

Example 13

PDT of lesions in a blood vessel

A targeted antibody-photosensitizer conjugate (APC) 160 is prepared using an antibody raised against antigens present on a lesion 164, where the lesion is of a type selected from the group consisting of atherosclerotic lesions, arteriovenous malformations, aneurysms, and venous lesions. Alternatively, a ligand-photosensitizer conjugate is prepared using a ligand that binds to a receptor protein 165 present on a lesion.

Please replace the paragraph on page 39, lines 10-14, with the following paragraph:

A variation of this method provides for the preparation of a conjugate 171 of a vessel wall 179 lesion 177 specific protein 173 or ligand 172 to a sonic energy 176 activated compound 174 and irradiated transcutaneously using and ultrasound probe 175 external to the skin 170. (See Fig. 17)

In the Claims:

Please replace claims 23, 24, 26, 27 and 30 with the following claims (a marked-up copy of the amended specification is attached to this Amendment):

23. (Amended) The method of claim 22, wherein said activation may be incrementally increased or decreased through the respective increase or decrease in irradiation intensity.

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24. (Amended) The method of claim 1, wherein said activation may be initiated through initiating irradiation or halted through discontinuing irradiation within a therapeutically reasonable time after the photosensitizing agent or prodrug has been administered but prior to biodegradation of said agent or prodrug.

26. (Amended) The method of claim 25, wherein said activation may be incrementally increased or decreased through the respective increase or decrease in irradiation intensity.

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27. (Amended) The method of claim 16, wherein said activation may be initiated through initiating irradiation or halted through discontinuing irradiation within a therapeutically reasonable time after the photosensitizing agent or prodrug has been administered but prior to biodegradation of said agent or prodrug.

30. (Amended) The method of claim 18, wherein said activation may be initiated through initiating irradiation or halted through discontinuing irradiation within a therapeutically reasonable time after the photosensitizing agent or prodrug has been administered but prior to biodegradation of said agent or prodrug.

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REMARKS

Any fees that may be due in connection with this application throughout its pendency may be charged to Deposit Account No. 50-1213.

The specification is amended to correct obvious typographical and spelling errors and to produce grammatical clarity. The amendment to the paragraph on page 13, lines 25-33, of the specification replaces the number "14" with the number —12—. The amendment finds basis in Figure 1 in which light has been labled as item number 12. The amendment to the paragraph beginning on page 31, line 22, through page 32, line 18, of the specification

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inserts the inadvertently omitted phrase —The APC— to render the sentence grammatically correct. The amendment finds basis at page 31, lines 28-32 which further discuss the APC. The amendment to the paragraph on page 34, lines 12-25, of the specification inserts the inadvertently omitted word "compound" for grammatical clarity. The amendment finds basis at page 33, lines 19-20, which discuss the compound. The amendment to the paragraph on page 36, lines 7-13, of the specification inserts the inadvertently omitted word —hours— for grammatical clarity. The amendment to the paragraph on page 39, lines 10-14, replaces the number "178" by the number —170—. The amendment finds basis in Figure 17 in which skin has been labled as item number 170.

Claims 23, 24, 26, 27 and 30 are amended to correct minor typographical errors.

No new matter has been added.

Included as an attachment is a marked-up version of the specification paragraphs and claims, per 37 CFR §1.121.

* * *

Entry of this amendment and examination of the application are respectfully requested.

Respectfully submitted,
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